

**A New Understanding of Ebola Virus Disease: A Review of Post Ebola Syndrome and Viral Persistence**

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## **Abstract**

Ebola Virus Disease (EVD), caused by Ebola virus, is highly fatal and is mostly restricted to African countries. EVD was previously described as an acute illness, but chronic illness can result as well, making infection more of a public health concern in recent years. New developments have demonstrated that individuals can have new and/or lingering symptoms post recovery, termed post-Ebola syndrome. Another phenomenon is the persistence of Ebola virus in immune privileged sites in survivors post recovery that can lead to viral transmission and/or relapsing infection.

Four cohort studies were reviewed that looked at symptoms in survivors, with different levels of follow-up and criteria. A total of 1,833 survivors were enrolled in these studies, all survivors of the 2014-2016 outbreak. This outbreak was very difficult to control, which led to more cases of disease and more disease survivors. Symptoms reported most frequently in survivors included joint pain, headache, and fatigue. Persistence of Ebola virus post recovery has most often been detected in semen but has also been found in breast milk and ocular fluid. Viral persistence in semen has led to a minimum of 112 cases of EVD. Breast milk was linked to at least one case of EVD (from mother to child), but ocular persistence has not yet been linked to transmission.

Ebola virus is a zoonotic pathogen, making eradication an unlikely scenario. There is increased need for prevention efforts to combat the high rates of post-Ebola syndrome and instances of viral persistence in survivors, including increasing vaccination efforts of the population at risk and receipt of EVD specific treatments in infected individuals. Treatments

specific for post-Ebola syndrome need to be assessed to help the growing population suffering from chronic symptoms. Despite the invaluable studies reviewed in this analysis, the pathogenesis and emergence of post-Ebola syndrome and EBOV persistence is relatively unknown and requires further study to make a meaningful impact on the 500 million individuals at risk for EVD in African countries.

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## **Preface**

This essay was made possible with the constant guidance and immense support of Dr. Jeremy Martinson. Dr. Kristen Mertz, my committee member, provided invaluable feedback during the essay process and I am so grateful for her assistance. I am so fortunate to have had my friends Kelly Schenk, Larisa Garza Chapa and Fiorella Nicoloso support me through this whole process. Lastly, I must thank my cousin, Rachel Lewack, my uncle, Steven Lewack and my grandpa, James Dailey (Pitt alum) for the unwavering support throughout my degree and essay process.

## **1.0 Introduction**

### **1.1 Introduction to Ebola Virus**

Ebola Virus (EBOV), a hemorrhagic fever virus, is the causative agent of Ebola Virus Disease (EVD). Ebola virus was discovered in 1976 near the Ebola River in the Democratic Republic of Congo, then Zaire. There are two main hemorrhagic fever viruses in the filovirus family, Ebola and Marburg, both with mortality rates up to 90%. Six species of ebolaviruses have been discovered: Zaire, Sudan, Bundibugyo, Tai Forest, Reston, and Bombali, but this review will only focus on Zaire ebolavirus henceforth referred to as Ebola virus/EBOV (Di Paola et al., 2020). EBOV has caused 28 outbreaks thus far, ranging from one case to almost 30,000 cases. Any reintroduction of the virus into the human population is considered an outbreak, no matter how few cases result (Centers for Disease Control and Prevention, 2023).

Until 2015, EVD was considered an acute illness, however, following the 2014-2016 outbreak of EVD in Western Africa, disease survivors began experiencing symptoms linked to their prior infection, termed post-Ebola syndrome (Wohl et al., 2023). This outbreak was the largest in EVD history with approximately 28,646 cases and 17,303 survivors. Guinea, Liberia, and Sierra Leone experienced the largest burden of morbidity and mortality, but the outbreak impacted multiple other countries including Mali, Nigeria, and the United States (Centers for Disease Control and Prevention, 2023). Although acute EVD is relatively well understood and documented, little is known about the impact of post-Ebola syndrome and viral persistence.

**Table 1: Cases and Deaths from EVD by Decade.**

	Cases	Deaths	Average Mortality Rate
1970s	318	281	88%
1980s	0	0	N/A
1990s	459	352	77%
2000s	545	466	86%
2010s	32,247	13,696	42%
2020s	182	88	48%

Currently, at least two studies have investigated symptom incidence in survivors and their un-infected contacts to examine the difference between symptoms related to EBOV infection and other causes (PREVAIL III study group, 2019; Bond et al., 2021). Two additional studies look at symptom incidence in survivors alone (Wohl et al., 2023; Tiffany et al., 2016). There is research to suggest that post-Ebola syndrome is not novel, but more evident as increasing numbers of people contract the virus and survive. As indicated in Table 1, there were only 223 known survivors of EVD prior to the 2010s, after which, there were over 18,000 survivors. Other factors involved in the recent detection of post-Ebola syndrome may include increased scientific understanding of EBOV, more robust research endeavors on EVD, and differences in transmissibility and virulence by disease-causing strain. While multiple studies explore different factors that may influence post-Ebola syndrome development, there is little research that provides a comprehensive overview of the existing literature.

At the same time post-Ebola syndrome was becoming evident, so was viral persistence in immune privileged sites in survivors. These discoveries coincided, likely as a result of the number of survivors from the 2014-2016 outbreak. Persistence of Ebola virus post recovery has mainly been noted in semen and breast milk, both with transmission potential. Viral persistence also has the potential to cause relapsing infection in the survivor, which is not required for the individual to transmit the virus. The mechanism(s) involved in persistence are largely unknown and need further research. Thus, the aim of this essay is to describe what is currently known about both post-Ebola syndrome and viral persistence, and where further research is needed.

## **1.2 Ebola Virus Epidemiology**

Ebola virus was originally introduced into the human population by zoonotic spillover, in which the virus is transmitted from an infected animal to a human. Transmission can occur through hunting, touching, butchering, or consumption of the infected animal or carcass. These spillover events are a major cause of EVD outbreaks. After initial human infection, the virus effectively spreads from person to person through direct contact with bodily fluids through broken skin and/or mucus membranes and through fomites (clothing, bedding, needles, medical equipment) that are contaminated by infectious bodily fluids (Jacob et al., 2020). The incubation period of EBOV ranges from 2-21 days, with an average of 9-10 days before symptoms begin after the virus has been introduced into the body (Baseler et al., 2017). The virus is not contagious until after symptom onset (World Health Organization, 2021).

Ebola virus can infect most human cells, but predominately targets mononuclear phagocytes (found in the liver, bone marrow, kidneys, and lungs) and dendritic cells. Innate

immune cells, once infected with EBOV, inadvertently contribute to viral dissemination by migrating from the site of infection to the lymph nodes, liver, and spleen. EBOV proteins VP35 and VP24 inhibit the cellular processes that aid in viral clearance and allow the virus to spread through the body. Macrophages, in response to EBOV, produce proinflammatory cytokines, causing an influx of macrophages to the site of infection which can result in the breakdown of endothelial barriers and hypovolemic shock via cytokine storm (Jacob et al., 2020).

Early symptoms of EVD are generally non-specific febrile symptoms (fever, headache, fatigue, arthralgia) followed by severe gastrointestinal symptoms (nausea, vomiting, diarrhea, abdominal pain). Later symptoms can include anemia, hemorrhage, hemolysis, hypoglycemia, mental status changes, renal failure, and uveitis (Baseler et al., 2017; Jacob et al., 2020). Due to viral persistence in semen samples observed after EBOV infection, men are instructed to practice safe sex or abstinence for at least one year after disease resolution or two negative semen samples (Jacob et al., 2020). However, the virus can persist in semen for as long as 40 months and individuals do not always have access to testing, therefore sexual transmission may be a contributing factor in disease spread (Di Paola et al., 2020). While sexual transmission from semen has been observed, female-to-male sexual transmission has not been identified in any cases (Subissi et al., 2018).

Pregnant people are at especially high risk when infected with EBOV as the virus can cross the placenta leading to infection and elevated risk of death in the fetus. In the rare event that a fetus is carried to term, they are likely to harbor the virus and are at elevated risk of death. EBOV RNA has been detected at high concentrations in amniotic fluid, the placenta, fetal tissue, and breast milk, indicating the multitude of ways a fetus and an infant can acquire infection from their mother. It is unknown how previous infection effects a survivor's later ability to carry a healthy fetus to

term, but it is hypothesized that there is a high risk of fetal death and babies born may harbor the virus (Jacob et al., 2020; Baseler et al., 2017). EBOV, therefore, may impact children born with the virus differently compared to individuals who acquired the virus post-birth. Thus, recognizing the specific risk of post-Ebola syndrome in both fetuses and babies may aid in understanding the different birth outcomes observed after an EVD outbreak.

Prior to 2020, there were no EVD specific treatments. Patients just received supportive care, which has been shown to significantly reduce mortality and has led to more disease survivors (Baseler et al., 2017). Supportive care can include intravenous fluids, blood infusions for hemorrhaging patients, pain management, and vasopressors to prevent hypovolemic/septic shock (Jacob et al., 2020). Vital signs should be continuously monitored to ensure appropriate supportive care is being provided (Baseler et al., 2017). In 2020, two monoclonal antibody treatments targeting EBOV glycoproteins (Inmazeb and Ebanga) were approved after undergoing a clinical trial during the 2018-2020 outbreak in the Democratic Republic of Congo (Jacob et al., 2020; World Health Organization, 2021). Inmazeb and Ebanga reduced mortality by 17% and 14% respectively in a randomized clinical trial, however, it is unknown if these treatments will lead to more EVD survivors long-term (Jacob et al., 2020). Two EBOV vaccines have been FDA approved, Ervebo and Zabdeno/Mvabea. Ervebo can be used pre- or post-exposure in an outbreak setting and has been shown to be effective in clinical trials. Zabdeno/Mvabea has relatively low efficacy and cannot be used in an outbreak setting as it requires two doses for protection. An effective, durable vaccine is invaluable, as half a billion individuals are at risk for Ebola virus infection (Woolsey and Geisbert, 2021).

### 1.3 Public Health Significance

Ebola virus outbreaks result in major strains on healthcare systems/infrastructure, making it more difficult to effectively treat a mass influx of patients (Woolsey and Geisbert, 2021). Outbreaks of EVD have varying case fatality rates, with a range of 25-88%. In general, later outbreaks have lower CFRs than the earlier outbreaks, but more cases (Centers for Disease Control and Prevention, 2023). Factors affecting the change in fatality rates may include increased viral transmissibility, decreased virulence, better case diagnostics resulting in fewer cases being missed, and advances in medicine leading to better survival rates and more individuals seeking medical care. This decrease in fatality, along with the increase in number of cases is additionally likely to be related to the occurrence of post-Ebola syndrome. Inadequate treatment resulting from lack of resources is likely also a contributing factor in later development of post-Ebola syndrome, which leads to high morbidity in survivors.

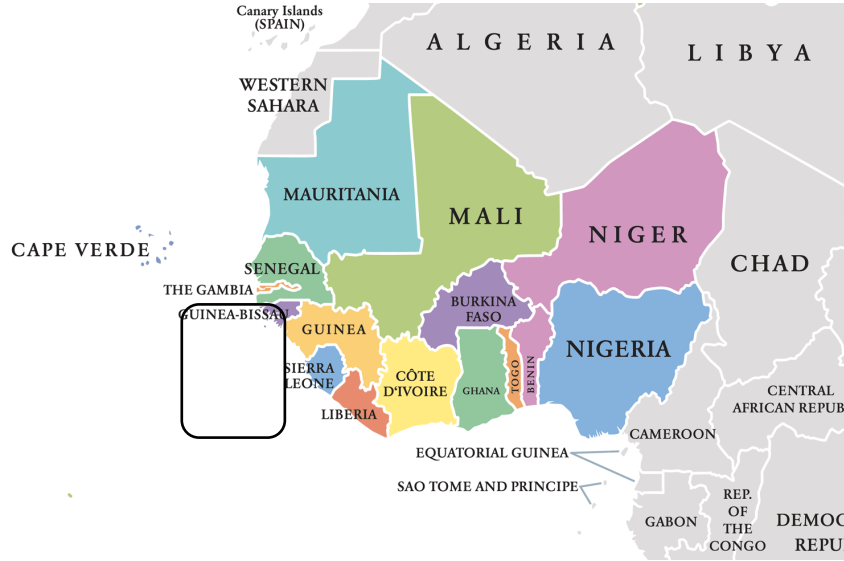
Individuals who have survived EVD have lasting emotional distress and post-traumatic stress disorder (PTSD). Survivors have not only experienced a life-threatening event themselves, but they have also witnessed other individuals dying from the disease, including family members. The inability to observe their traditional burial practices contributes to depression both in survivors and un-infected family members. One study found that 71% of survivors surveyed reported symptoms of PTSD including rapid heart rate, abdominal discomfort and dizziness when reminded of their experience one month post discharge from an Ebola Treatment Unit (Jacob et al., 2020). The role of post-Ebola syndrome on the psyche of survivors has not been fully accessed but is likely a contributing factor to psychiatric disorders, and symptoms resulting have been compared to those seen in combat veterans with PTSD (PREVAIL III study group, 2019; Tiffany et al., 2016).

## **2.0 Methodology and Methods**

A literature search was conducted using google scholar and PubMed using keywords including “EVD,” “Post Ebola Syndrome,” “Persistence of Ebola virus,” “Post Ebola Syndrome studies,” and “Pathogenesis.” Papers were excluded if they were written in a foreign language. Original search parameters limited papers by publication date (articles written before 2019 were excluded) to review the most current information before widening the search to look for earlier mentions of post Ebola syndrome and viral persistence. Approximately sixty papers were reviewed, and fourteen were chosen. Papers regarding post-Ebola syndrome and viral persistence were chosen based on study size and information provided (larger studies provided more information than multiple case studies in this instance). Papers included date back as far as 2015.



### 3.0 Ebola Virus Outbreaks and Outbreak Epidemiology



**Figure 1: Map of Western Africa.** Highlighting displays the countries where the 2014-2016 outbreak began. Source: Shopify Imagery.

The West African Epidemic, the largest EVD outbreak in history, was most likely the result of a spillover event. The outbreak began in a forested region of Guinea in March of 2014 and quickly spread to both Liberia and Sierra Leone, countries bordering Guinea (Centers for Disease Control and Prevention, 2023). As shown in Figure 1, these three countries are very close together with minimal borders between them. Guinea, Sierra Leone, and Liberia were grossly unprepared for a disease outbreak, having recently recovered from years of civil unrest leading to severe damage to their already poor health infrastructure. Additionally, these three countries are among the poorest in the world, further contributing to their lack of preparedness and massive uncontrolled spread of disease throughout the area (World Health Organization, 2015). These three

countries alone accounted for an estimated 28,610 cases of EVD during this outbreak, with a case fatality rate of 39% (Centers for Disease Control and Prevention, 2023).

**Table 2: Outbreaks of EVD (50 Cases or More) by Year and Location.**

<b>Outbreak Year and Location</b>	<b>Number of Cases</b>	<b>Number of Deaths</b>	<b>Case Fatality Rate (%)</b>	<b>Outbreak Cause</b>
<b>Democratic Republic of Congo: 1976</b>	318	280	88%	Spillover
<b>Gabon: 1994</b>	51	31	61%	Unreported
<b>Democratic Republic of Congo: 1995</b>	315	254	81%	Spillover*
<b>Gabon: 1996</b>	60	45	75%	Spillover*
<b>Gabon: 2001</b>	65	53	81%	Spillover*
<b>Republic of Congo: 2001</b>	59	44	75%	Spillover*
<b>Republic of Congo: 2003</b>	143	128	89%	Spillover
<b>Democratic Republic of Congo: 2007</b>	264	187	71%	Unreported
<b>West African Epidemic: 2014-2016</b>	28,616	11,310	39%	Spillover
<b>Guinea</b>	3,814	2,544	66.7%	
<b>Sierra Leone</b>	14,124	3,956	28%	
<b>Liberia</b>	10,678	4,810	45%	
<b>Democratic Republic of Congo: 2014</b>	69	49	71%	Spillover*
<b>Democratic Republic of Congo: 2018</b>	54	33	61%	Unreported
<b>Democratic Republic of Congo: 2018-2020</b>	3,470	2,287	66%	Unreported

<b>Democratic Republic of Congo: 2020</b>	130	55	42.3%	Spillover + Persistent infection
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\*Indicates hypothesized spillover by CDC or based on description indicating contact with wildlife or index case near forested area

Recently, at least four outbreaks have been linked via sequencing to persistent infections in survivors from earlier outbreaks. It is nearly impossible to definitively determine the exact source of an outbreak, even with sequencing of patient samples, therefore sources are hypotheses or theories ascertained by the CDC based on collected data. Table 2 shows causes of outbreaks in which there were 50 or more cases of disease. Early outbreaks with unreported causes are likely spillovers, but recent outbreaks can be the result of spillover or persistent infection. In 2020, 54 cases of EVD were linked to persistent infection in a survivor from the 2018 outbreak in the Democratic Republic of Congo (other cases of this outbreak resulted from spillover). Two smaller outbreaks in the Democratic Republic of Congo in 2021 were linked via persistence to the 2018-2020 outbreak. In 2021, an outbreak in Guinea resulted from viral persistence in a survivor of the 2014-2016 outbreak—at least four years later (Centers for Disease Control and Prevention, 2023).

The link between post-Ebola syndrome and viral persistence remains unknown, as only one of the outbreaks caused by persistence was related to the West African Epidemic, where post-Ebola syndrome has mostly been described. The mechanism allowing Ebola virus to both persist in the body after an individual has tested negative for the virus and to reactivate and cause EVD is unknown. One theory is that the virus can travel from the area in which it is latent in the body to an area that it can replicate again and cause disease. Areas of potential viral persistence include the central nervous system, eyes, testes, and breast milk (Di Paola et al., 2020; World Health Organization, 2021).

## 4.0 Post Ebola Syndrome

After the 2014-2016 Ebola Virus Outbreak, many individuals reported symptoms believed to be related to their prior infection with EBOV, termed post-Ebola syndrome. Post Ebola syndrome refers to the ongoing health complications and abnormal symptoms observed in individuals that have recovered from Ebola virus infection. Two studies with large cohorts have been conducted to compare symptoms between individuals who have recovered from EVD and their household contacts to study post-Ebola syndrome (Bond et al., 2021; PREVAIL III study group, 2019). Two other studies observed symptom incidence, but only in EVD survivors (Wohl et al., 2023; Tiffany et al., 2016). Studies conducted by *Bond et al.* and *Tiffany et al.*, focused on individuals in Sierra Leone, while the PREVAIL III Study Group and *Wohl et al.* study focused on individuals in Liberia (Bond et al., 2021; PREVAIL III Study Group, 2019; Wohl et al., 2023; Tiffany et al., 2016). These studies, along with others conducted on the subject, demonstrate that patients who survive EVD infection can have ongoing health complications because of infection (PREVAIL III Study Group, 2019; Wohl et al., 2023).

The PREVAIL III study group enrolled 966 EVD survivors and 2,350 of their close contacts, enrolling up to nine contacts per survivor. On average, baseline visits for survivors occurred 358 days post EVD onset. Participants were followed for one year with information collected at three intervals: baseline, six months, and twelve months. Most participants remained enrolled throughout the study (90%), some individuals passed away during the study and others were lost to follow-up. Examination of participants included past, interim, and current medical history data collection, current symptom questionnaire, physical examination, and blood

collection. Eye examinations were completed in a subset of study participants (PREVAIL III Study Group, 2019).

Seven symptoms recorded during this study were reported in survivors at statistically significant levels ( $p < 0.01$ ) when compared to the control group: urinary frequency, headache, fatigue, muscle pain, memory loss, joint pain, and muscle tenderness. Uveitis was observed at high levels in both survivors and contacts, but the incidence of uveitis was significantly higher in survivors. Survivors had higher levels of abnormal findings of statistical significance on abdominal, chest, neurologic, muscle, and joint exams. No differences between the groups were observed in renal, hepatic, or hematopoietic measurements. Symptoms decreased after baseline visits for both groups. Approximately 85% of survivors and 11% of contacts were positive for Ebola virus antibodies. These positive contacts had lower antibody titers than survivors. Of this small portion of contacts, 47% reported symptoms indicative of EVD around the time their related survivor was infected. Of contacts negative for EBOV antibodies, 31% reported previous symptoms consistent with EVD infection. These two observations indicate limitations of the study, as it is likely that a portion of individuals classified as contacts (without previous EVD infection) had unreported EVD (PREVAIL III Study Group, 2019).

The *Bond et al.* study enrolled 375 EVD survivors and 1,040 of their household contacts. Patient symptoms were determined via questionnaire, allowing for potential recall bias, followed by physical examinations. Survivors were enrolled at least two years after disease resolution. Patients were asked about constitutional, musculoskeletal, ophthalmologic, cardiac, gastrointestinal, psychiatric, neurologic, and auditory symptoms by category. Reported symptoms included joint tenderness, decreased range of motion, hallucinations, difficulty sleeping, chest pain, heart palpitations, tinnitus, blurry vision, excessive fatigue, loss of appetite, and dizziness.

At least 70% of EVD survivors reported at least one new symptom and over 50% reported symptoms in two or more categories. Constitutional (fever, headache) and musculoskeletal symptoms were seen in the highest numbers in EVD survivors in general, with the most reported symptoms being joint pain and headache (Bond et al., 2021).

The *Wohl et al.* study enrolled 326 survivors with a median of 389 days post-acute EVD and accessed eight cardinal symptoms: headache, fatigue, joint pain, muscle pain, hearing loss, visual loss, numbness in hands and numbness in feet. Participants were followed for approximately six years, with all but 24 participants remaining for the entire period. Study visits were conducted every three months for 18 visits, then every six months where participants were administered a symptom survey in which they described symptom incidence and severity by Likert scale. Severity was determined by the level of interference in the participant's life. Symptoms characterized occurred post Ebola Treatment Unit discharge. Data presented was collected from June of 2015 to March of 2022. This cohort did not have access to vaccines or EVD treatments, just supportive care. During the study period, severity of six of the eight symptoms declined during each follow-up period, with no statistically significant change in numbness of hands and feet. Of the participants sampled, 245 individuals reported at least one or more symptoms at baseline, with 161 participants still reporting at least one symptom five years later. The most common symptoms reported were joint pain, headache, and fatigue, with joint pain rated as most interfering in the participants' lives. Based on COVID-19 research, it is theorized that viral persistence in immune-privileged sites may be a source of antigen that stimulates the immune response and triggers symptoms, indicating that viral persistence and post-Ebola syndrome are highly related (Wohl et al., 2023).

The *Tiffany et al.* study group enrolled 166 survivors for their clinic where clinical and psychological assessments were conducted at each visit via medical examination, symptom questionnaire and psychiatric screening. Patients visited weekly, then bimonthly/monthly until symptom resolution, with an average of 4.3 follow-up visits per patient. Free symptom treatment was offered, with referral to other centers when needed. The most frequent symptoms observed in this cohort were arthralgia, fatigue, abdominal pain, headache, anemia, skin disorders, back pain, alopecia, and ocular complications (mainly uveitis). Arthralgia was the most common symptom, reported by 78% of patients, followed by fatigue at 70%. Of psychiatric symptoms, 30 patients (18%) reported insomnia, five (3%) reported hallucinations and four (2.4%) reported irritability. Of considerable note, one woman became pregnant during follow-up and delivered a healthy baby. Additional theories for post-Ebola syndrome described in this study include immune complex deposition, persistence of Ebola virus antigen and molecular mimicry (Tiffany et al., 2016). Invaluable information was gained from all studies, but the pathogenesis of post-Ebola syndrome remains widely uncertain and requires further study (PREVAIL III Study Group, 2019).

## 5.0 Viral Persistence

Another phenomenon observed post West African Epidemic is the persistence of EBOV RNA in immune privileged sites, mainly the eyes and testes, that can lead to relapsing EVD. *Mbala-Kingebeni et al.* conducted a case report on a 25-year-old individual who survived an initial Ebola virus infection, but later became ill with the virus again as a result of persistence and promoted person to person spread of EBOV. This patient originally presented with EVD in June of 2019, even after having received Ervebo vaccination six months prior. The patient was treated with monoclonal antibodies (Ebanga) and recovered, resulting in discharge from the ETU two weeks after admission (after EBOV RNA was no longer detectable). Four months later, EVD symptoms presented in the patient, and he returned to the ETU eight days later after a short hospital stay. The following day, the patient showed signs of acute kidney and liver injury and soon developed acute respiratory distress before falling into a coma and dying (Mbala-Kingebeni et al., 2021).

Epidemiological investigation determined that this patient directly caused 29 cases of EVD and indirectly caused an additional 62 cases. Both this patient's initial and persistent infection occurred during the 2018-2020 outbreak of EBOV that took place in the Democratic Republic of Congo (DRC). Sequencing of EBOV RNA from patient samples determined that this patient's second infection with EBOV was the result of viral reactivation, rather than from outward viral transmission. Two main questions arise from this patient's case: why did he develop EVD after vaccination and why did relapsing infection occur in an otherwise healthy individual? Theories include that IgG titers against the virus did not develop after vaccination, the virus was able to escape immune detection while it remained persistent in the patient and/or that the virus remained



persistent in an immune-privileged site of the body, such as the eye. Approximately 20% of individuals who received the same vaccine did not develop IgG binding titers against the virus. Additionally, researchers found no evidence of conditions resulting in immunocompromise or a failed antibody response in this patient. To understand both why and how Ebola virus becomes persistent in the body after recovery from infection, further data analysis is needed (Mbalakinge et al., 2021).

One study examined eight instances in which viral persistence resulted in the spread of disease (Subissi et al., 2018). These instances could not be linked to circulating Ebola virus in the region, making transmission from ongoing chains highly unlikely. Four of these instances can be definitively linked to sexual transmission via persistence in semen. The transmission route is unknown in three of these cases. Of special note, the last of these cases is that of a nine-month-old that tested positive for EBOV post-mortem. Both parents were likely undiagnosed EVD survivors, with positive IgG results for Ebola virus antigen. The mother's breast milk was positive for EBOV, the most likely route of transmission of infection to the infant, and the father's semen was positive for EBOV. These infections caused by viral persistence led to an additional 23 total cases of EVD (Subissi et al., 2018).

Another interesting case is that of a 43-year-old physician that contracted EVD while treating patients in Sierra Leone and was found to have viral persistence in the aqueous humor of the eye at least nine weeks after clearance of viremia, i.e., blood tests negative for EBOV (Varkey et al., 2015). Semen samples collected were also positive for EBOV. Treatment for EVD infection had included an experimental small interfering RNA antiviral, convalescent plasma, and supportive care. His only lingering symptoms from infection were difficulties with word-finding and exercise intolerance. Ophthalmic symptoms began shortly after discharge from Emory

University Hospital, where he was treated, including photophobia and a burning/foreign body sensation in the eyes. One month later, multiple scars with halos were found in both eyes and an intraretinal hemorrhage in the left eye upon examination resulting in a diagnosis of posterior uveitis. Uveitis in the left eye continued to get worse for three months before improvement. Conjunctival swabs and tears were collected for testing, and both were negative for EBOV. The treating physicians believe that the ocular symptoms were a result of direct cytopathic effect of active replication of Ebola virus in the eye (Varkey et al., 2015).

In addition to studying symptom incidence to understand post-Ebola syndrome, the PREVAIL III study group completed semen sampling for a subset of their survivor cohort to look at viral persistence. Semen sampling was completed for 267 male survivors. Of these individuals, 81 had EBOV RNA detected at least once and 252 individuals provided more than one sample. Detection of viral RNA was intermittent for 78 individuals, with 36 men testing negative twice prior to a positive result. Persistence in semen does not always indicate presence of infectious virus and no association was found between the level of antibody present in blood or semen and the symptoms experienced (PREVAIL III study group, 2019).

## 6.0 Discussion

There are four possible outcomes of EVD: death, clearance of the virus but morbidity from post-Ebola syndrome, clearance of the virus from the blood but persistence in immune privileged sites that can lead to transmission and/or relapsing infection and complete clearance of the virus with no ill effects (the least common). Unfortunately, there are still a lot of unknowns regarding post Ebola syndrome and persistence of Ebola virus RNA, but the studies discussed describe novel information that greatly contributes to the knowledge in these areas. Some of these unknowns that require future research include: (1) how is the virus able to persist in immune privileged sites in the body and how does this only occur in some individuals, (2) how does the virus reactivate in the body to cause relapsing infection, (3) how is the virus able to cause transmission when present in immune privileged sites when the surviving individual is not actively infected, and (4) how related are post-Ebola syndrome and viral persistence—i.e., do symptoms correlate with where Ebola virus/antigen is remaining in the body.

Ebolaviruses are difficult to study due to their high fatality rates (since they must be studied in a high-level laboratory equipped for hazardous agents and handled only by highly trained professionals) and the inability to adequately mimic disease progression in animal models. Additionally, as EVD primarily impacts low-income, resource poor countries, there is less of an urgency among high-income countries to investigate the disease (Jacob et al., 2020).

Prevention of EVD begins with reducing contact with wildlife, which can prevent spillover events from occurring, thus preventing the virus from re-entering the human population. Appropriate personal protective equipment must be worn when caring for patients in both community and healthcare settings. Additionally, measures should be taken to avoid transmission

from survivors that may have virus persisting in immune privileged sites, including practice of safe sex and additional care taken by healthcare providers during examinations (World Health Organization, 2021).

There are currently no therapeutics targeted for treating post-Ebola syndrome and studies in this regard are lacking. Patients presenting with post-Ebola syndrome only receive symptom management, with no attempt at treating the cause of the symptoms. Two directed treatment options should be considered for these patients: vaccination with Ervebo and receipt of monoclonal antibody treatments (Inmazeb or Ebanga). The goal of vaccination is to activate antibody production to help quell any persisting virus that may be causing the chronic symptoms. Monoclonal antibody treatment could help to reduce symptom severity in these patients. Research on these treatments is needed to determine efficacy.

As of April 2023, there are no current outbreaks of EVD. The most recent outbreak of EVD occurred in the Democratic Republic of Congo in 2022 and only resulted in one case of disease (Centers for Disease Control and Prevention, 2023). As Ebola virus is a zoonotic pathogen, it is almost impossible to eradicate. Therefore, there will continue to be outbreaks of EVD in African countries, with half a billion individuals at risk.

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